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14. ABSTRACT The proposal centers on developing the principal investigator (PI) into an independent prostate cancer physician-scientist, using as a vehicle this DoD award with specific research aims to examine the ERG oncoprotein as a target for prostate cancer therapy by using novel transgenic mice. As many as 50% of prostate cancers possess a chromosomal translocation involving the <i>ERG</i> oncogene. I hypothesized that ERG can serve as an effective molecular therapeutic target for prostate tumors using novel prostate tumor mouse models. During this fifth year of support we have not been able to adhere to our "Statement of Work" – for <u>Task#2</u> or <u>Task#3</u> . We were successful at completing <u>Task#1</u> , but characterization of ERG expression from our prostate mouse model did not demonstrate any detectable prostate specific ERG expression at the protein level. To remedy this issue, we re-started <u>Task #1</u> two years ago with the new prostate specific TET driver mouse, <i>Hoxb13-rtTA</i> . We have spent this last year examining whether ERG can collaborate with <i>AKT1</i> with these new mice, <i>Hoxb13-rtTA/tetO-ERG</i> . We had to reinitiate breeding of more mice using a different <i>tetO-ERG</i> founder line and are in the midst of processing samples for analysis. Despite these setbacks, concurrently during this award period and made possible by this DoD award, the PI has made significant strides in promoting his career as an independently funded prostate cancer physician-scientist with national and international recognition.					
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Evaluating the efficacy of ERG targeted therapy in vivo for prostate tumors

PI – Phuoc T. Tran, MD, PhD

1. INTRODUCTION:

The proposal centers on developing the principal investigator (PI) into an independent prostate cancer physician-scientist, using as a vehicle this DoD award with specific research Aims to examine the ERG oncoprotein as a target for prostate cancer therapy by using novel transgenic mice. Prostate cancer is the most common cancer diagnosed in men in the United States. It has been estimated that greater than 200,000 new cases of prostate cancer were diagnosed in the United States in 2012 and prostate cancer was responsible for ~30,000 deaths or the second most common cause of cancer deaths in men (1). Recent efforts to classify distinct molecular subtypes of prostate cancer have led to the novel findings that greater than 50% of prostate cancers possess a chromosomal translocation involving the *ETS* oncogene family of transcription factors (2, 3). These *ETS* translocations result in dysregulated overexpression of the *ETS* oncogene in prostate cancer cells. The most common *ETS* family member involved in these translocation events is the v-ets erythroblastosis virus E26 oncogene homolog (ERG). Most molecular targeted therapies in other cancers are notable for their lack of serious side-effects and amazing tolerability. I hypothesized that *ERG*, the most common *ETS* oncogene found to be mutated in prostate cancer can serve as an effective molecular therapeutic target for prostate tumors. I planned to show this with novel autochthonous prostate tumor mouse models. I also hypothesized that *ERG* facilitates tumorigenesis alone or in the context of activated AKT1 by dysregulating proliferation, apoptosis and/or senescence programs *in vivo*. Demonstrating whether prostate tumors in mouse models are dependent for *ERG* for tumor survival would be the first proof of principle demonstration of molecularly targeted therapy for spontaneously arising prostate tumors *in living* animals. Ultimately, this mentored award has the goal of protecting the research time of the PI to allow development of his research program so that he may become a future leader in prostate cancer research.

The original specific aims are below:

Specific Aim#1 - Generate and characterize an inducible *ERG* prostate specific mouse model.

Rationale: I have created a novel prostate TET system mouse model and am interested in the effects of *ERG* expression alone and in combination with *AKT1* in the prostate.

Study Design: I will validate inducible expression of both *ERG* and *Luc* *in vivo* using real time-RT-PCR (qPCR), BLI of whole living animals and by organ Western analysis in bi-transgenic *ARR2PB-tTA/ ERG-tetO-Luc* (AE) mice.

Specific Aim#2 – Determine if *ERG* cooperates with *AKT1* for prostate tumorigenesis.

Rationale: *ERG* overexpression *in vitro* suggests that *ERG* may facilitate tumorigenesis, but *ERG* transgenic mouse models vary in the severity of their tumor phenotypes alone and with *AKT1* co-overexpression. The mechanism for *ERG* prostate phenotypes alone or in combination with *AKT1* overexpression *in vivo* are unknown.

Study Design: Generate *ARR2PB-tTA/MPAKT1/ ERG-tetO-Luc* (AA1E) tri-transgenic mice and compare to single oncogene mice to genetically analyze cooperation *in vivo*. Investigate using molecular techniques if *ERG* modulates proliferation, apoptosis and/or senescence programs *in vivo*.

Specific Aim#3 - Determine if *ERG* can serve as an effective molecular therapeutic target for prostate tumors *in vivo*.

Rationale: Despite the importance that *ERG* overexpression is believed to play in prostate tumorigenesis, the therapeutic value of targeting *ERG* on autochthonous prostate tumors has not been tested *in vivo*. The mechanism for any autochthonous tumor regression or stasis *in vivo* upon *ERG* inactivation is unknown.

Study Design: Following development of autochthonous prostate tumors in TET regulated mice I will treat mice with doxycycline to simulate targeted treatment against the *ERG* oncogene. Investigate using molecular techniques if *ERG* inactivation modulates proliferation, apoptosis and/or senescence programs in autochthonous prostate tumors *in vivo*.

2. KEYWORDS:

ERG

Prostate cancer

Inducible transgenic mouse model

3. OVERALL PROJECT SUMMARY:

Progress is listed in relation to each specific task in the “Statement of Work” and highlighted by *italics* for Years 1-4 and **BOLD** font for the past year (Year 5).

Task#1 - Generate and characterize an inducible *ERG* prostate specific mouse model (months 1-17).

Numbers of mice surviving weaning and for mating: 65

1a. IACUC and other regulatory approval process for animal work (months 1-4).

As reported in our Year 1-4 Progress Reports, we applied for and obtained approval from the Johns Hopkins Sidney Kimmel Comprehensive Cancer Center IACUC for the studies described in our DoD grant award (see Appendix for documentation approval).

We had to get re-approval of our IACUC protocol this past year (see Appendix)

1a. Mating mice to characterize (months 4-10).

As reported in our Year 1-4 Progress Reports, the appropriate single transgene ARR2PB-tTA (A) and ERG-tetO-Luc (E) mice were mated to produce cohorts of (AE) bitransgenic mice. There were no issues with producing the required numbers of AE mice. We stated in the last progress report (Year 4) that we were generating more bitransgenic, Hoxb13-rtTA/ERG-tetO-luc (HE) and tritransgenic animals, Hoxb13-rtTA/MPAKT1/ERG-tetO-luc (HA1E) mice to perform Task #1 and Task #2, respectively.

We have characterized these HE mice using as a reporter, functional expression of luc detected with bioluminescence imaging (BLI) and molecular characterization with qPCR and Western for ERG (data not shown; see Table 1B and Table 2B). We did not see robust BLI signal from the HE mice, nor have we observed high level of ERG expression at the mRNA or protein levels (data not shown). This was in contrast to what we observed with a very similar line Twist1-tetO-luc crossed to the prostate specific TET driver Hoxb13-rtTA (4) (see Figure 1 Appendix). Similarly, we have shown that another founder line of ERG-tetO-luc can express luc from other tissue specific drivers such as those that drive expression ubiquitously (see Figure 2 Appendix) or to the liver (data no shown). The possible explanations are that the HE transgenic combination we selected was not compatible or the particular tetO-ERG founder line we used may have undergone silencing which is a well characterized phenomenon in transgenic mice. We switched to a different tetO-ERG founder line and began to generate more bitransgenic, Hoxb13-rtTA/ERG-tetO-luc (HE) and tritransgenic Hoxb13-rtTA/MPAKT1/ERG-tetO-luc (HA1E) mice to perform Task #1 and Task #2, respectively

1b. Collecting tissues from AE mice to characterize ERG expression (months 8-14). AE mice will be weaned and placed on water without doxycycline and 5 males for each of the following age time points: 4, 8, 12 and 24 weeks (n=25 mice total, 5 additional for incidentals), will be interrogated using the assays mentioned below in 1d.

As reported in our Year 1-4 Progress Reports, the appropriate numbers of AE bitransgenic mice (n=25) had been placed on drinking water without doxycycline to activate the ERG transgene.

We collected tissues from HE mice with and without doxycycline (Table 1B). We will proceed with this sub-Task #1b again when we get more animals with a new founder tetO-ERG line (see Task #1a above).

1c. Collecting tissues from AE mice turned OFF to characterize inducible ERG expression (months 8-14). 12 week old males will be followed for the OFF time points: 1, 2 and 4 weeks (n=20 mice total, 5 additional for incidentals) and tissues extracted for interrogation using the assays mentioned below in 1d.

As reported in our Year 1-4 Progress Reports, the appropriate numbers of AE bitransgenic mice have been placed on regular water (n=20) for 4-6 weeks following weaning to activate the *ERG* transgene followed by changing to doxycycline drinking water (0.2 mg/ml) changed weekly to inactivate the *ERG* transgene. **We collected tissues from HE mice turned OFF (Table 2B). We will proceed with this sub-Task #1c again when we get more animals with a new founder tetO-*ERG* line (see Task #1a above).**

1d. Performing experiments on tissues from mice (months 14-17). Tissues from 1b and 1c above will be harvested for histology and flash frozen for molecular studies: prostate lobes, other genitourinary (GU) organs, lungs, heart, liver and spleen. These specimens will then be processed for H&E histology and immunohistochemistry (IHC) performed using anti-Myc, anti-FLAG and anti-luciferase antibodies to confirm prostate luminal cell epithelia expression. Whole lobe and organ Western blotting using the same antibodies will also be performed and transcription of *ERG* confirmed with specimens using qPCR. See Table 1 and 2 below for summary of results. We were able to harvest AE mice as above for all the “ON” time points at least 5 mice: 4, 8, 12 and 24 weeks. Similarly, for the “OFF” time points we have been able to collect tissues from ≥ 5 mice from the 1, 2 and 4 week time points.

We do not see robust BLI signal from the HE mice, nor have we observed high level of *ERG* expression at the mRNA or protein levels (data not shown & Tables 1B and 2B below).

We have performed analysis as summarized below in Table 1A & 2A. The AE mice from the “ON” time points collected have had no abnormalities on gross or H&E examination of their prostates. The other organs in these mice (lungs, heart, liver and spleen) were also normal on necropsy. Similarly, the AE mice from the “ON” and “OFF” time course displayed no pathology on gross or histologic exam of the H&E slides. We have attempted IHC and westerns for protein expression of *ERG* that is tagged by Myc and FLAG epitope tags, but have not been able to see expression using either approach. We also attempted on a limited scale luc IHC and *ERG* qPCR with these samples which were similarly negative.

We performed some analysis on the HE mice as summarized below in Table 1B & 2B and we do not observe any gross abnormalities in any of the samples. Nor did we observe any expression of *ERG* by BLI or other molecular techniques.

1e. Analyzing results of experiments on tissues from mice (months 14-17). See Table 1 and Table 2 for summary of results and “Conclusions” below for explanation of results.

Table 1A – Summary of Task #1b & d to date.

Genotype	4 wks On DOX	8 wks On DOX	12 wks On DOX	24 wks On DOX
AE	6 mice	7 mice	5 mice	5 mice
Gross	WNL	WNL	WNL	WNL
Histologic	WNL	WNL	WNL	WNL
Myc IHC	Negative expression	Negative expression	Negative expression	Negative expression
FLAG IHC	Negative expression	Negative expression	Negative expression	Negative expression
luc IHC	ND	ND	ND	Negative expression
FLAG Western	Negative expression	Negative expression	ND	Negative expression
<i>ERG</i> qPCR	ND	ND	ND	Negative expression

A – *ARR2PB-tTA*; DOX – doxycycline; E – *luc-tetO-ERG*; IHC – immunohistochemistry; qPCR – quantitative polymerase chain reaction; WNL – within normal limits.

Table 1B – Summary of Task #1b & d to date.

Genotype	4 wks On DOX	8 wks On DOX	12 wks On DOX	24 wks On DOX
HE	4 mice	6 mice	6 mice	5 mice
Gross	WNL	WNL	WNL	ND
Histologic	WNL	WNL	WNL	ND
Myc IHC	Negative expression	Negative expression	ND	ND
FLAG IHC	Negative expression	Negative expression	ND	ND
luc IHC	ND	ND	ND	ND
FLAG Western	Negative expression	Negative expression	ND	ND
ERG qPCR	Negative expression	Negative expression	ND	ND

A – *ARR2PB-tTA*; DOX – doxycycline; E – *luc-tetO-ERG*; IHC – immunohistochemistry; qPCR – quantitative polymerase chain reaction; WNL – within normal limits.

Table 2A – Summary of Task #1c & d to date.

Genotype	1 wks Off DOX	2 wks Off DOX	4 wks Off DOX
AE	6 mice	6 mice	6 mice
Gross	WNL	WNL	WNL
Histologic	WNL	WNL	WNL
Myc IHC	Negative expression	ND	ND
FLAG IHC	Negative expression	ND	ND
luc IHC	Negative expression	ND	ND
FLAG Western	Negative expression	ND	ND
ERG qPCR	Negative expression	ND	ND
IHC	Negative expression	ND	ND
Western	Negative expression	ND	ND

A – *ARR2PB-tTA*; DOX – doxycycline; E – *luc-tetO-ERG*; IHC – immunohistochemistry; qPCR – quantitative polymerase chain reaction; WNL – within normal limits; ND - not done.

Table 2B – Summary of Task #1c & d to date.

Genotype	1 wks Off DOX	2 wks Off DOX	4 wks Off DOX
HE	3 mice	3 mice	ND
Gross	WNL	WNL	ND
Histologic	WNL	WNL	ND
Myc IHC	ND	ND	ND
FLAG IHC	ND	ND	ND
luc IHC	ND	ND	ND

FLAG Western	ND	ND	ND
ERG qPCR	ND	ND	ND
IHC	ND	ND	ND
Western	ND	ND	ND

A – *ARR2PB-tTA*; DOX – doxycycline; E – *luc-tetO-ERG*; IHC – immunohistochemistry; qPCR – quantitative polymerase chain reaction; WNL – within normal limits; ND - not done.

Many of the steps/tasks below are dependent on the steps above and have not been initiated.

Task#2 - Determine if *ERG* cooperates with *AKT1* for prostate tumorigenesis (months 14-34).

Numbers of mice surviving weaning and for mating: 150

2a. Mating mice for cooperation experiments (months 14-20).

We bred *Hoxb13-rtTA/MPAKT1/ERG-tetO-Luc* (HA1E) mice for Task #2a, but aborted and sacrificed these specific HA1E mice for the reasons as explained above in Task #1d. We have re-initiated the mating required to produce *Hoxb13-rtTA/MPAKT1/ERG-tetO-Luc* (HA1E) mice using a new founder tetO-ERG line.

2b. Collecting tissues from cooperation experiments (months 18-30).

2c. Performing experiments on tissues from mice (months 20-32). Tissues from 2b above will be harvested for histology and flash frozen for molecular studies: prostate lobes, other GU organs, lungs, heart, liver and spleen. These specimens will then be processed for H&E histology and IHC performed using anti-Myc, anti-FLAG and anti-luciferase antibodies. Whole lobe and organ Western blotting using the same antibodies will also be performed and transcription of *ERG* confirmed with specimens using qPCR. IHC for cleaved caspase 3 (CC3) and Ki-67. Senescence markers such as p15, p16, p21 and p27 will be analyzed by IHC and qPCR. In addition, I will perform senescence associated beta-galactosidase (SA- β -gal) staining.

2d. Analyzing results of experiments on tissues from mice (months 22-34).

Each of the steps/tasks below is dependent on the steps above and has not been initiated.

Task#3 - Determine if *ERG* can serve as an effective molecular therapeutic target for prostate tumors *in vivo* (months 34-60)

Numbers of mice surviving weaning and for mating: 120

3a. Mating mice for therapeutic experiments (months 34-40).

3b. Collecting tissues from therapeutic experiments mice ON 6-12 months and then OFF 1-6 months (months 40-56).

3c. Performing experiments on tissues from mice (months 42-58). Tissues from 3b above will be harvested for histology and flash frozen for molecular studies: prostate lobes, other GU organs, lungs, heart, liver and spleen. These specimens will then be processed for H&E histology and IHC performed for Myc, FLAG, luciferase, CC3, Ki-67, p15, p16, p21 and p27. Whole lobe and organ Western blotting using the same antibodies will also be performed and transcription of *ERG* confirmed with specimens using qPCR. In addition, I will perform SA- β -gal staining.

3d. Analyzing results of experiments on tissues from mice (months 44-60).

4. KEY RESEARCH ACCOMPLISHMENTS:

- *Confirmation that our ARR2Pb-tTA mouse line is not robust enough to drive expression of tetO-regulated genes in the mouse prostate.*

- Characterization of the HE transgenic mouse showed that either the transgene combination used was not compatible or this particular tetO-ERG founder line we used may have undergone silencing which is a well characterized phenomenon in transgenic mice.
- Re-initiated breeding for novel bitransgenic, *Hoxb13-rtTA/ERG-tetO-Luc* (HE) and tritransgenic animals, *Hoxb13-rtTA/MPAKT1/ERG-tetO-Luc* (HA1E), using the more robust prostate specific driver *Hoxb13-rtTA* and a new founder tetO-ERG line.

5. CONCLUSION:

During this fifth and last year of support we have not been able to adhere to the timeline of our “Statement of Work” - Task#2 - Determine if *ERG* cooperates with *AKT1* for prostate tumorigenesis (months 14-34) or Task#3 - Determine if *ERG* can serve as an effective molecular therapeutic target for prostate tumors *in vivo* (months 34-60). We have been previously successful at completing the tasks for Task#1 - Generate and characterize an inducible *ERG* prostate specific mouse model (months 1-17), but this characterization of *ERG* expression from our old prostate inducible mouse model, *ARR2PB-tTA*, did not demonstrate any detectable prostate specific *ERG* expression at the protein level using Western or IHC (see Tables 1A & 2A above).

We had in our Year 2 progress report concluded that the lack of a prostate phenotype despite prostate epithelium specific expression of other tetO reporter lines was due to the low level of expression from the *ARR2PB-tTA* line and perhaps insufficient for the *in vivo* experiments described in our proposal.

In Year 3 we attempted to remedy this issue with low prostate specific expression and proposed to re-start Task #1 of the project with the new prostate specific TET driver mouse, *Hoxb13-rtTA* (H) (4), in collaboration with Dr. Charles Bieberich. The *Hoxb13-rtTA* line allows for much more robust expression of tetO target genes as compared to our original *ARR2PB-tTA* line (see Figure 1 Appendix).

In Years 3-4, the breeding between our tetO-ERG mice and Dr. Bieberich’s *Hoxb13-rtTA* mice had been problematic, but we overcame these issues and were able to proceed with Tasks #1a-c with the *Hoxb13-rtTA/tetO-ERG* (HE) mice (see Table 1B & 2B). We also started breeding mice for Task #2a, but aborted and sacrificed these specific HA1E mice for the reasons below.

In this final Year 5, we completed Task #1d with the HE mice and continued with breeding HA1E mice for Task #2 studies. Unfortunately, we did not observe expression with BLI, via qPCR for *ERG* or Western for *ERG* in HE mice (see Table 1B & 2B). We have troubleshot this situation and believe the specific HE transgenic mouse combination we used was not compatible or this particular tetO-ERG line we used may have undergone silencing which is a well characterized phenomenon in transgenic mice. We are in the process of breeding with a new founder tetO-ERG line more HE mice (Task #1a) to proceed again with Tasks #1b-d.

Finally, the ultimate goal of this DoD Prostate Cancer Physician Research Training Award (PRTA) was to help develop the PI into an independent prostate cancer researcher. Concurrently during this entire award period and made possible by this DoD award, the PI has made significant strides in promoting his career as an independently funded prostate cancer physician-scientist with national and international recognition.

“So What”

Despite the importance that *ERG* overexpression is believed to play in prostate tumorigenesis, the therapeutic value of targeting *ERG* rearrangements has not been tested *in vivo*. The ability to interrogate using *in vivo* model systems whether *ERG* or other oncogenes are good molecular therapeutic targets could provide a huge leap forward for prostate cancer research and treatment of prostate cancer patients. Demonstrating whether prostate tumors in my inducible transgenic mice are dependent for *ERG* for tumor maintenance would be the first proof of principle demonstration of molecularly targeted therapy for prostate tumors *in vivo* and we will be able to determine whether molecularly targeted therapy against *ERG* in the context of activated *AKT1* would be an effective therapy for prostate tumors.

The ultimate goal of this DoD PRTA was to develop the PI into an independent prostate cancer researcher. The PI has made significant strides in promoting his career as an independently funded prostate cancer physician-scientist with national and international recognition.

6. PUBLICATIONS, ABSTRACTS AND PRESENTATIONS:

- During this fifth and last year of support we have not published any manuscripts, abstracts or presented work directly from the Aims proposed in this DOD PRTA at any venue other than at our own private lab meetings. However, in the spirit of this award protecting the research time of the PI, this has enabled our group to contribute the following reportable outcomes related to prostate cancer research:

1. Lay Press:

We were awarded a Movember-PCF Challenge grant to test a novel radiation and immunotherapy combination in oligometastatic prostate cancer patients.

1. http://www.pcf.org/site/c.leJRIOrEpH/b.9303981/k.F89E/6_New_Movember_Foundation_8211_PCF_Challenge_Awards_Advance_Precision_Medicine_and_Novel_Treatments_for_Lethal_Prostate_Cancer.htm
2. <http://www.prnewswire.com/news-releases/6-new-movember-foundation--pcf-challenge-awards-advance-precision-medicine-and-novel-treatments-for-lethal-prostate-cancer-300122883.html>
3. <http://ir.advaxis.com/press-releases/detail/1174/advaxiss-adxs-psa-awarded-research-grants-from-the-prostate-cancer-foundation-and-the-movember-foundation>

We were noticed for a donation to our prostate cancer research efforts by a local philanthropic group.

4. <http://www.severnaparkvoice.com/community/local-ravens-fans-combine-passion-football-and-charity>

Coverage of a collaborative study where we developed a novel small animal optical imaging platform for preclinical research.

5. <http://medicalphysicsweb.org/cws/article/research/64139>

2. Peer-Reviewed Scientific Journals (Since the beginning of the DoD PRTA):

1. **Phuoc T. Tran**⁺, Russell K. Hales, Jing Zeng, Khaled Aziz, Tarek Salih, Rajendra P. Gajula, Sivarajan Chettiar, Nishant Gandhi, Aaron T. Wild, Rachit Kumar, Joseph M. Herman, Danny Song and Theodore L. DeWeese. Tissue Biomarkers for Prostate Cancer Radiation Therapy. ***Curr Mol Med*** 12 (2012) 772-787. PMID: 22292443; PMCID: PMC3412203.
+ - corresponding author.
2. Nishant Gandhi*, Aaron T. Wild*, Sivarajan T. Chettiar, Khaled Aziz, Yoshinori Kato, Rajendra P. Gajula, Russell D. Williams, Jessica Cades, Anvesh Annadanam, Danny Song, Yonggang Zhang, Russell K. Hales, Joseph M. Herman, Theodore L. DeWeese, Edward M. Schaeffer, **Phuoc T. Tran**. Novel Hsp90 inhibitor NVP-AUY922 radiosensitizes prostate cancer cells. ***Cancer Biol Ther*** 14 (2013) 347-356. PMID: 23358469. PMCID: PMC3667875.
* - these authors contributed equally.
3. Jason A. Efstathiou, Deborah S. Nassif, Todd R. McNutt, C. Bob Bogardus, Walter Bosch, Jeff Carlin, Ron C. Chen, Henry Chou, Dave Eggert, Benedick Fraass, Joel Goldwein, Karen E. Hoffman, Ken Hotz, Margie Hunt, Marc Kessler, Colleen A. F. Lawton, Chuck Mayo, Jeff M. Michalski, Sasa Mutic, Louis Potters, Chris M. Rose, Howard M. Sandler, Greg Sharp, Wolfgang Tomé, **Phuoc T. Tran**, Terry Wall, Anthony L. Zietman, Peter E. Gabriel, Justin E. Bekelman. Practice-Based Evidence to Evidence-Based Practice: Building the National Radiation Oncology Registry. ***J Oncol Pract*** 9 (2013) e90-95. PMID: 23942508. PMCID: PMC3651578.
4. Sara Alcorn*, Amanda J. Walker*, Nishant Gandhi, Amol Narang, Aaron T. Wild, Russell K. Hales, Joseph M. Herman, Danny Y. Song, Theodore L. DeWeese, Emmanuel Antonarakis, **Phuoc T. Tran**. Molecularly Targeted Agents as Radiosensitizers in Cancer Therapy – Focus

on Prostate Cancer. *Int J Mol Sci* 14 (2013) 14800-14832. PMID: 23863691. PMCID: PMC3742274.

* - these authors contributed equally.

5. Rajendra P. Gajula*, Sivarajan T. Chettiar*, Russell D. Williams, Saravanan Thiagarajan, Yoshinori Kato, Khaled Aziz, Ruoyi Wang, Nishant Gandhi, Aaron T. Wild, Farhad Vesuna, Jinfang Ma, Tarek Salih, Jessica Cades, Elana Fertig, Shyam Biswal, Timothy F. Burns, Christine Chung, Charles M. Rudin, Joseph M. Herman, Russell K. Hales, Venu Raman, Steven An, **Phuoc T. Tran**. The twist box domain is required for Twist1-induced prostate cancer metastasis. *Mol Cancer Res* 11 (2013) 1387-1400**. PMID: 23982216. PMCID: PMC3833995.
* - these authors contributed equally.
** - Cover illustration and Highlighted in *Mol Cancer Res*.
6. Debasish Sundi, Vinson Wang, Phillip M. Pierorazio, Misop Han, Alan W. Partin, **Phuoc T. Tran**, Ashley E. Ross, Trinity J. Bivalacqua. Identification of men with the highest risk of early disease recurrence after radical prostatectomy. *Prostate* 74 (2014) 628-36. PMID: 24453066; PMCID: PMC4076164.
7. Minh-Phuong Huynh-Le, Zhe Zhang, **Phuoc T. Tran**, Theodore L. DeWeese, Daniel Y. Song. Low inter-rater reliability in grading of rectal bleeding using NCI-CTC and RTOG toxicity scales: a survey of radiation oncologists. *Int J Radiat Oncol Biol Phys* 90 (2014) 1076-1082. PMID: 25442040. PMCID: PMC4276525.
8. Rajendra P. Gajula*, Sivarajan T. Chettiar*, Russell D. Williams, Katriana Nugent, Yoshinori Kato, Hailun Wang, Reem Malek, Kekoa Taparra, Jessica Cades, Anvesh Annadanam, A-Rum Yoon, Elana Fertig, Beth A. Firulli, Lucia Mazzacurati, Timothy F. Burns, Anthony B. Firulli, Steven An, **Phuoc T. Tran**. Structure-function studies of the bHLH phosphorylation domain of Twist1 in prostate cancer cells. *Neoplasia* 17 (2015) 16-31. PMID: 25622896. PMCID: PMC4309734.
* - these authors contributed equally.
9. Lynnette R Ferguson, Helen Chen, Andrew R. Collins, Marisa Connell, Giovanna Damia, Santanu Dasgupta, Meenakshi Malholtra, Alan K Meeker, Amedeo Amedei, Amr Amin, S. Salman Ashraf, Katia Aquilano, Asfar S. Azmi, Dipita Bhakta, Alan Bilsland, Chandra S. Boosani, Sophie Chen, Maria Rosa Ciriolo, Hiromasa Fujii, Gunjan Guha, Dorota Halicka, William G. Helferich, W. Nicol Keith, Sulma I. Mohammed, Elena Niccolai, Xujuan Yang, Kanya Honoki, VirginiaR. Parslow, Satya Prakash, Sarallah Rezazadeh, Rodney E Shackelford, David Sidransky, **Phuoc T Tran**, Eddy S. Yang, and Christopher A Maxwell. Genomic instability in human cancer: molecular insights and opportunities for therapeutic attack and prevention through diet and nutrition. *Semin Cancer Biol* 35 (2015) S5-S24. PMID: 25869442. PMCID: PMC4600419.
10. Keith I. Block,...**Phuoc T Tran**,... Massimo Zollo. Designing a broad-spectrum integrative approach for cancer prevention and treatment. *Semin Cancer Biol* 35 (2015) S276-S304. PMID: 26590477. PMCID: PMC4819002.
11. Emelyn H. Shroff, Livia S. Eberlin, Vanessa M. Dang, Arvin M. Gouw, Meital Gabay, Stacey J. Adam, David I. Bellovin, **Phuoc T. Tran**, William M. Philbricke, Adolfo Garcia-Ocanaf, Stephanie C. Casey, Yulin Li, Chi V. Dang, Richard Zare, Dean W. Felsher. MYC Oncogene Overexpression Drives Renal Cell Carcinoma in a Mouse Model through Glutamine

Metabolism. Proc Natl Acad Sci USA 112 (2015) 6539-44**. PMID: 25964345. PMCID: PMC4450371.

** - Highlighted in *Cancer Res – Breaking Advances*.

12. Wolfgang Lilleby*, Amol Narrang*, Gunnar Tafjord, Ljiljana Vlatkovic, Kjell Magne Russnes, Andreas Stensvold, Knut Håkon Hole, **Phuoc Tran**, Karsten Eilertsen. Favorable outcomes in locally advanced and node positive prostate cancer patients treated with combined pelvic IMRT and androgen deprivation therapy. Radiat Oncol 10 (2015) 232. PMID: 26577452. PMCID: PMC4650510.

* - these authors contributed equally.

13. Andrew J. Armstrong, Susan Halabi, Patrick Healy, W. Robert Lee, Bridget F. Koontz, Judd W. Moul, Kelly Mundy, Patricia Creel, Sarah Yenser Wood, Kristen Davis, Michael A. Carducci, Mark N. Stein, Brooke Reimer, Avery N. Spitz, Minh Nguyen, Ellen Bratt, Sung Kim, **Phuoc T. Tran**, Daniel J. George. A phase 2 multimodality trial of docetaxel/prednisone with sunitinib followed by salvage radiation therapy in men with PSA recurrent prostate cancer after radical prostatectomy. Prostate Cancer Prostatic Dis 19 (2016) 100-106. PMID: 26754260.

14. Amol K. Narang, Carol Gergis, Scott P. Robertson, Pei He, Ashwin N. Ram, Todd R. McNutt, Emily Griffith, Tate DeWeese, Stephanie Honig, Harleen Singh, Daniel Y. Song, **Phuoc T. Tran**, Theodore L. DeWeese. Very-high-risk localized prostate cancer: outcomes following definitive radiation. Int J Radiat Oncol Biol Phys 94 (2016) 254-262. PMID: 26853334.

15. Bin Zhang, Ken Kang-Hsin Wang, Jingjing Yu, Sohrab Eslami, **Phuoc T. Tran**, Iulian Iordachita, Michael S. Patterson, John W. Wong. Bioluminescence tomography guided radiation therapy for preclinical research. Int J Radiat Oncol Biol Phys 94 (2016) 1144-53**. *In press*. PMID: 26876954. PMCID: PMC4814325.

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(<http://medicalphysicsweb.org/cws/article/research/64139>).

16. Steven P. Rowe, Michael A. Gorin, Mohamad E. Allaf, Kenneth J. Pienta, **Phuoc T. Tran**, Martin G. Pomper, Ashley E. Ross and Steve Y. Cho. PET Imaging of the Prostate-Specific Membrane Antigen in Prostate Cancer – Current State of the Art and Future Challenges. Prostate Cancer Prostatic Dis (2016). *In press*.

17. Ashley Ross, Robert Den, Kasra Yousefi, Bruce J. Trock, Jeffrey Tosoian, Elai Davicioni, Darby J. S. Thompson, Voleak Choerung, Zaid Haddad, **Phuoc T. Tran**, Edward J. Trabulsi, Leonard G. Gomella, Costas D. Lallas, Firas Abdollah, Felix Y. Feng, Eric A. Klein, Adam P. Dicker, Stephen J. Freedland, R. Jeffery Karnes, Edward M. Schaeffer. Efficacy of Post-Operative Radiation in a Prostatectomy Cohort Adjusted for Clinical and Genomic Risk. Prostate Cancer Prostatic Dis (2016). *In press*.

18. Kekoa A. Taparra, **Phuoc T. Tran**, Natasha Zachara. Hijacking the hexosamine biosynthetic pathway to promote epithelial-mesenchymal transition-mediated neoplastic phenotypes. Front Oncol (2016). *In press*.

19. Reem Malek*, Hailun Wang*, Kekoa Taparra, **Phuoc T. Tran**. Therapeutic targeting of epithelial plasticity programs – Focus on the epithelial-mesenchymal transition. Cells Tissues Organs (2016). *Conditionally accepted*.

* - these authors contributed equally.

3. Invited Articles (Since the beginning of the DoD PRTA):

1. **Phuoc T. Tran**, Trinity J. Bivalacqua, Adam P. Dicker. Adjuvant Radiation for Node Positive Disease Post-Prostatectomy - More Good News But Who Will Listen? ***J Clin Oncol*** 32 (2014) 3917-3919. PMID: 25311219.
2. **Phuoc T. Tran**, Trinity J. Bivalacqua, Adam P. Dicker. Reply to CG Rusthoven et. al. ***J Clin Oncol*** 33 (2015) 1990-1. PMID: 25847932.
3. Amol K. Narang and **Phuoc T. Tran**. Prostate cancer: Case volume and improved outcomes across cancer care. ***Nat Rev Urol*** 13 (2016) 186-7. PMID: 26832160. PMCID: PMC4821678.
4. Jeffrey J. Tosoian, **Phuoc T. Tran**, Ashley E. Ross. Concurrent Androgen Deprivation with Radiotherapy – A Cautionary Tale of “Do As I Say, Not As I Do”? ***Eur Urol*** (2016). *In press*. PMID: 27080744.

4. Abstracts (Year 5 only from a total of 47 since the beginning of the DoD PRTA):

1. Omar Y. Mian, Scott Robertson, Amol Narang, Hee Joon Bae, **Phuoc Tran**, Theodore L. DeWeese, Danny Y. Song. A Retrospective Review of 366 Patients Treated With LDR Prostate Brachytherapy at a Single Institution: Histopathologic, Biochemical, and Dosimetric Predictors of Outcome. American Brachytherapy Society (ABS) 2015 National Meeting.
2. Omar Y. Mian, Scott Robertson, Amol Narang, Sameer Agarwal, Hee Joon Bae, Todd McNutt, **Phuoc Tran**, Theodore L. DeWeese, Danny Y. Song. Dosimetric predictors of sexual function decline following LDR brachytherapy for prostate cancer (PCa). ASTRO 2015 National Meeting.
3. Ken Kang-Hsin Wang, Bin Zhang, **Phuoc T. Tran**, Iulian Iordachita, Michael S. Patterson, and John W. Wong. Bioluminescence tomography guided system for small animal radiation research platform (SARRP). **Oral presentation** for the SPIE Photonics West 2016.
4. Ashley Ross, Robert Den, Kasra Yousefi, Bruce Trock, Elai Davicioni, Darby Thompson, Voleak Choerung, Zaid Haddad, Jeff Tosoian, **Phuoc Tran**, Firas Abdollah, Felix Feng, Adam Dicker, Stephen Freedland, R. Jeffery Karnes, Edward Schaeffer. Efficacy of Early and Delayed Radiation in a Prostatectomy Cohort Adjusted for Genomic and Clinical Risk. SUO Annual Meeting 2015.
5. Benjamin Benzon, Stephanie A. Glavaris, Brian W. Simons, Robert M. Hughes, Rebecca M Miller, Katriana Nugent, Brian Shinder, Lee Blosser, **Phuoc Tran**, Paula J Hurley, Milena Vuica-Ross, Edward M. Schaeffer, Charles G. Drake, Ashley E. Ross. Effect of local therapy on the systemic anti-tumor response in prostate cancer. SUO Annual Meeting 2015.
6. Benjamin Benzon, Mandeep Takhar, Nicholas Erho, Mohammed Alshalalfa, Paula Hurley, Stephanie Glavaris, Brian Simons, Michael Haffner, **Phuoc Tran**, Felix Feng, Elai Davicioni, Jeffrey Karnes, Edward M. Schaeffer, Charles G. Drake, Ashley E. Ross. B7-H3 expression is androgen related and predictive of prostate cancer outcomes in a large natural history cohort of men undergoing prostatectomy. SUO Annual Meeting 2015.
7. Jeffrey J. Tosoian, Debasish Sundi, Meera Chappidi, Ridwan Alam, Kenneth J. Pienta, **Phuoc T. Tran**, Edward M. Schaeffer, Ashley E. Ross. Contemporary Treatment Patterns and Short-Term Outcomes in Men with Very High Risk Prostate Cancer. SUO Annual Meeting 2015.

8. Robert Den, Kasra Yousefi, Bruce J. Trock, Elai Davicioni, Jeffrey Tosoian, Darby J. S. Thompson, Voleak Choeurng, Zaid Haddad, **Phuoc T. Tran**, Edouard John Trabulsi, Leonard G. Gomella, Costas D. Lallas, Firas Abdollah, Felix Yi-Chung Feng, Adam Dicker, Stephen J. Freedland, Jeffrey Karnes, Edward M. Schaeffer, Ashley Ross. Efficacy of Early and Delayed Radiation in a Prostatectomy Cohort Adjusted for Genomic and Clinical Risk. Genitourinary Cancers Symposium 2016.
9. Ashley Ross, Benjamin Benzon, Shuang Zhao, Mandeep Takhar, Michael Haffner, Nicholas Erho, Paula Hurley, Jeffrey J. Tosoian, Mohammed Alshalalfa, Stephanie Glavaris, Brian Simons, **Phuoc T. Tran**, Elai Davicioni, Jeffrey Karnes, Edward M. Schaeffer, Charles G. Drake, Felix Y. Feng. The Relationship of B7H3 expression to androgen and prostate cancer outcomes in a large natural history cohort of men undergoing prostatectomy. Genitourinary Cancers Symposium 2016.
10. Ashley Ross, Benjamin Benzon, Stephanie Glavaris, Brian Simons, Robert Hughes, Patrick Mullane, Rebecca Miller, Katriana Nugent, Brian Shinder, Richard Blosser, **Phuoc T. Tran**, Paula Hurley, Milena Vuica-Ross, Edward M. Schaeffer, Charles G. Drake. Effect of local therapy on the systemic anti-tumor response in prostate cancer. Genitourinary Cancers Symposium 2016.
11. Omar Y Mian, Scott P. Robertson, Amol Narang, Sameer Aggarwal, Hee Joon Bae, Carol Gergis, Todd R. McNutt, **Phuoc T. Tran**, Theodore L. DeWeese, Danny Song. Dosimetric predictors of sexual function decline following LDR brachytherapy for prostate cancer (PCa). Genitourinary Cancers Symposium 2016.
12. Michael A. Gorin, Steven P. Rowe, Margarita Mana-ay, Zsolt Szabo, Edward M. Schaeffer, **Phuoc T. Tran**, Mohamad E. Allaf, Curtiland Deville, Steve Y. Cho, Kenneth James Pienta, Martin G. Pomper, Ashley Ross. Study of PSMA-targeted ¹⁸F-DCFPyL PET/CT in the Evaluation of Men with an Elevated PSA Following Radical Prostatectomy. Genitourinary Cancers Symposium 2016.
13. Jeffrey J. Tosoian, Debasish Sundi, Brian Francis Chapin, Emmanuel S. Antonarakis, Meera Chappidi, Ridwan Alam, Stephanie Glavaris, R. Jeffrey Karnes, Kenneth James Pienta, **Phuoc T. Tran**, Edward M. Schaeffer, Ashley Ross. Contemporary Treatment Patterns and Short-Term Outcomes in Men with Very High Risk Prostate Cancer. Genitourinary Cancers Symposium 2016.
14. Ken Kang-Hsin Wang, Bin Zhang, **Phuoc T. Tran**, Iulian Iordachita, Michael S. Patterson, and John W. Wong. Bioluminescence tomography guided system for small animal radiation research platform (SARRP). 3rd Small Animal Precision Image-Guided Radiotherapy Symposium 2016.
15. Ashley E. Ross, Brian Shinder, Jeffrey Tosoian, Nicholas Erho, Mohammed Alshalalfa, Kasra Yousefi, Paula Hurley, Felix Feng, Elai Davicioni, **Phuoc Tran**, Edward Schaeffer. Primary Tumor Androgen Receptor Signaling as a Predictor of Castrate Resistance. AUA Annual Meeting 2016.
16. Meera R. Chappidi, Max Kates, Zeyad Schwen, Nikolai Sopko, **Phuoc Tran**, Nita Ahuja, Stephanie Terezakis, Phillip M. Pierorazio, Trinity J. Bivalacqua. Intraoperative Radiation Therapy During Genitourinary Surgery: The Importance of Specimen Margin Status in Improving Survival. AUA Annual Meeting 2016.

17. Jeffrey J. Tosoian, Debasish Sundi, Brian Chapin, R. Jeffrey Karnes, Emmanuel S. Antonarakis, Meera Chappidi, Ridwan Alam, Stephanie Glavaris, Kamyar Ghabili, Mohamad E. Allaf, Trinity J. Bivalacqua, Kenneth J. Pienta, **Phuoc T. Tran**, Edward M. Schaeffer, Ashley E. Ross. Trends in Surgical Management of High-Risk Prostate Cancer: Evidence of an Evolving Treatment Paradigm. AUA Annual Meeting 2016.
18. Jeffrey J. Tosoian, Debasish Sundi, Brian Chapin, R. Jeffrey Karnes, Emmanuel S. Antonarakis, Meera Chappidi, Ridwan Alam, Stephanie Glavaris, Kamyar Ghabili, Mohamad E. Allaf, Trinity J. Bivalacqua, Kenneth J. Pienta, **Phuoc T. Tran**, Edward M. Schaeffer, Ashley E. Ross. Predictors of Surgical Cure in Men with Very High Risk Prostate Cancer. AUA Annual Meeting 2016.
19. Russell D. Williams, Rajendra P. Gajula, Reem Malek, Belinda Nghiem, Katriana Nugent, Brian W. Simons, A-Rum Yoon, Hailun Wang, Kekoa Taparra, Lawrence True, Steven An, Danny Song, Theodore L. DeWeese, Edward M. Schaeffer, Kenneth J. Pienta, Paula J. Hurley, Colm Morrissey, **Phuoc T. Tran**. A TWIST1-MLL-WDR5-*HOTTIP* complex regulates *HOXA9* chromatin to facilitate metastasis of prostate cancer. 9th Multi-institutional Prostate Cancer SPORE Retreat.

SEMINARS/TALKS (Since the beginning of the DoD PRTA):

1. World Presidents' Organization Health Network Foundation Program for JHU Men's Health Day (November 20, 2010). "Prostate Cancer: Prevention, Screening and Treatment Options".
2. RTOG Semi-annual Meeting - Genitourinary Translational Research Program (January 14, 2011). "MYC as a biomarker to direct statin targeted radiosensitization for definitive treatment of prostate cancer".
3. George O'Brien Center at Johns Hopkins University Advisory Committee Meeting (January 25, 2011). "TWIST1 and Embryonic Reawakening in benign prostatic hyperplasia revisited".
4. JHU, Brady Urology Prostate Cancer Research Day (February 25, 2012). "Using High-Dose Statins to Target MYC-overexpressing Prostate Cancers".
5. JHU, Brady Urology Prostate Cancer Advisory Board Meeting (June 5, 2012). "Using High-Dose Statins to Target MYC-overexpressing Prostate Cancers".
6. I Congress of Oncology D'Or (Rio de Janeiro, Brazil) – Meeting with Johns Hopkins Experts (July 6, 2013). "Extreme Hypofractionation for Localized Prostate CA: Radiobiologic Rationale & Early Results".
7. Stanford University Medical Center, Radiation Oncology Visiting Professor (October 28, 2013). "Structure-functions studies of the TWIST1 oncoprotein in lung and prostate cancer".
8. Baltimore-Philadelphia Prostate Cancer Summit (November 1, 2013). "Phase I Trial of HSP90 inhibition and radiation-androgen deprivation therapy for high-risk, localized and locally advanced prostate cancer".
9. 6th Biennial TEMTIA Meeting: Symposium I – Cell/Molecular Biology of EMT (November 13, 2013). "The Twist box domain is required for TWIST1-induced metastasis of prostate cancer cells".
10. JHU, Brady Urology Prostate Cancer Research Day (February 8, 2014). "Phase I Trial of HSP90 inhibition and radiation-androgen deprivation therapy for high-risk, localized and locally advanced prostate cancer".
11. UC San Diego, Moores Cancer Center (April 18, 2014). "Structure-functions studies of the TWIST1 oncoprotein in lung and prostate cancer".
12. Delaware Society for Clinical Oncology (May 22, 2014). "Emerging genetic tests for localized prostate cancer: ready for prime time?".
13. Prostate Cancer UK 11th Biennial Prostate Cancer Forum (June 13, 2014). "SBRT/SBAR for Oligomet".

14. Amtrak Alliance - Baltimore-Philadelphia Prostate Cancer Summit (November 7, 2014). "SBRT/SBAR for Oligometasts".
15. JHU, SKCCC Translational Research Conference (February 11, 2015). "Credentialing TWIST1 as a Therapeutic Target in Lung and Prostate Cancer".
16. JHU, Brady Urology Prostate Cancer Working Group (February 20, 2015). "Consolidative Local Therapies for Oligometastatic Prostate Cancer".
17. JHU, Radiation Oncology Grand Rounds (February 24, 2015). "Credentialing TWIST1 as a Therapeutic Target in Lung and Prostate Cancer".
18. 8th Multi-institutional Prostate Cancer SPORE Retreat (March 16, 2015). "Stereotactic Ablative Radiation for Treatment of Oligometastatic Disease".
19. Georgetown University Medical Center, Biochemistry and Molecular & Cellular Biology (March 31, 2015). "Credentialing TWIST1 as a Therapeutic Target in Lung and Prostate Cancer".
20. PCF-Young Investigator Community Radiotherapy/ Radiobiology Working Group webinar (May 14, 2015 – Baltimore, MD). "SBRT for Oligometastatic Prostate Cancer".
21. AbbVie JHU Meeting (May 14, 2015 – Baltimore, MD). "AbbVie Visit: Credentialing TWIST1 & "Pre-clinical Models.
22. JHU 38th Biennial Meeting & Reunion – SKCCC Seminar (June 5, 2015 – Baltimore, MD). "Altering the Natural History of Oligometastatic Prostate Cancer with Local Ablative Therapies".
23. PCF Norway Prostate Cancer Symposium 2015 – Accelerating Innovations and Advancing Discovery (June 12, 2015 – Oslo, Norway). "Advanced Radio-therapeutic approaches to manage Prostate Cancer – Stereotactic Ablative Techniques for Oligometastatic Prostate Cancer".
24. PCF Norway Prostate Cancer Symposium 2015 – Accelerating Innovations and Advancing Discovery (June 12, 2015 – Oslo, Norway). "Advanced Radio-therapeutic approaches to manage Prostate Cancer - Proton Beam - Does this costly technology improve outcomes?".
25. PCF Coffey-Holden Prostate Cancer Academy Meeting (June 28, 2015 – La Jolla, CA). "Metastasis Directed Therapy – The Radiation Oncologist's Perspective".
26. JHU, Brady Urology Grand Rounds (September 10, 2015 – Baltimore, MD). "Stereotactic Ablative Techniques for Oligometastatic Prostate Cancer".
27. Movember Foundation Visiting Professor (October 13, 2015 – Melbourne, Australia). "Stereotactic Ablative Techniques for Oligometastatic Prostate Cancer".
28. Clemenceau Medical Center - Cancer Forum (October 30, 2015 – Beirut, Lebanon). "Oligometastatic disease: paradigm shift from palliative approach to curative approach".
29. University of Chicago, Radiation & Cellular Oncology (February 12, 2016 – Chicago, IL). "Metastatic Prostate Cancer – Basic to Clinical Interrogation".

7. INVENTIONS, PATENTS AND LICENSES:

Nothing to report.

8. REPORTABLE OUTCOMES:

Nothing to report.

9. OTHER ACHIEVEMENTS:

CLINICAL TRIALS:

1. J0910 - Multimodality Therapy for Recurrent High Risk Prostate Cancer: A Phase II Study. Role: PI. *Closed early.*
2. J1153 - Pharmacodynamic Trial of Pre-Prostatectomy Lovastatin on MYC Down-Regulation in Localized Prostate Cancer. Role: PI. *Closed early.*
3. J11157 - Stereotactic Body Radiation Therapy and Short-Term Androgen Ablation for Intermediate-Risk, Localized, Adenocarcinoma of the Prostate. Role: PI. *Open to accrual.*

4. J1454 - SALVENZA Trial: Phase II Randomized Placebo-Controlled Double-Blind Study of Salvage Radiation Therapy (SRT) Plus Placebo *Versus* SRT Plus Enzalutamide in Men with High-Risk PSA-Recurrent Prostate Cancer after Radical Prostatectomy. Role: PI. *Open to accrual.*
5. J1569 - Phase I Trial of HSP90 Inhibition and Radiation-Androgen Deprivation Therapy for High-Risk Localized and Locally Advanced Prostate Cancer. Role: PI. *Closed early.*
6. J15180 - Phase II Randomized Observation versus Stereotactic Ablative Radiation for Oligometastatic Prostate Cancer (ORIOLE) trial. Role: PI. *Open to accrual.*
7. J15203 - Pilot Biomarker Study of Stereotactic Body Radiation Therapy for Prostate Cancer Oligometastases. Role: PI. *Open to accrual.*
8. URO-004 - A Retrospective Study of Prolaris® for the Prediction of Progression in Men Treated with Modern External Beam Radiation Therapy for Prostate Cancer. Role: PI. *Under IRB review.*

GRANTS:

No additional funding was applied for based on this work specifically resulting from the proposed Aims. However, in the spirit of this award protecting the research time of the PI, this has enabled our group to apply for additional funding related to prostate cancer research:

CURRENT:

1. 1U01CA183031-01A1 Pomper/DeWeese (PI) 5/15/2015-3/31/2017
NIH/NCI
“PSMA-Directed PET/MR Imaging and Image-Guided Therapy of Prostate Cancer”
The overall goal is to validate a positron-emitting, PSMA-targeted imaging agent clinically so it may be used to full advantage in supporting existing and emerging therapies for a spectrum of patients suffering from prostate cancer.
Role: Co-I
2. ENZA-13L21 Tran/Antonarakis (PI) 3/2/2015-1/1/2020
Astellas-Medivation Pharma
“SALV-ENZA - Phase II Randomized Placebo-Controlled Double-Blind Study of Salvage Radiation Therapy (SRT) Plus Placebo vs. SRT Plus Enzalutamide in Men with High Risk PSA-Recurrent Prostate Cancer”
Randomized, double-blind, phase II, prospective, multicenter study in male adults with biochemically recurrent prostate cancer following radical prostatectomy.
Role: co-PI
3. Movember-PCF Challenge Tran (PI) 8/1/15-7/30/2017
Movember-Prostate Cancer Foundation (PCF)
“Altering the Natural History of Metastatic Prostate Cancer using Stereotactic Ablative Radiotherapy (SABR) and Immune Stimulation”
The overall goal is to test consolidation of all sites of macroscopic disease with SABR in combination with the immune stimulatory agent ADXS-PSA in men with oligometastatic prostate cancer using a first-in-man clinical trial and complimentary correlative approaches.
Role: PI
4. Patient-Centered Outcomes Res Smith (PI) 11/1/15-10/31/20
Patient-Centered Outcomes Research Institute (PCORI)
Simplifying Survivorship Care Planning: Comparing the Efficacy and Patient-Centeredness of Three Care Delivery Models in Prostate, Breast and Colorectal Cancer
Role: Co-I

COMPLETED:

- | | | |
|---|---------------------|------------------|
| 1. O'Brien Center Pilot | Tran/Schaeffer (PI) | 11/1/10-10/31/12 |
| George M. O'Brien Center for Benign Prostate Hyperplasia/LUTS Research | | |
| "TWIST1 and Embryonic Reawakening in benign prostatic hyperplasia revisited" | | |
| Aim #1 is to establish a link between prostate luminal cell specific <i>Twist1</i> overexpression and increased prostate stem cells. Aim #2 is to determine if prostate specific <i>Twist1</i> overexpression <i>in vivo</i> results in autochthonous prostate hyperplasia. | | |
| Role: PI | | |
| 2. PCW Award | Tran (PI) | 4/1/11-3/31/13 |
| Patrick C. Walsh Prostate Cancer Research Fund | | |
| "MYC as a biomarker to direct statin targeted therapy for definitive treatment of prostate cancer" | | |
| Aim #1: Pharmacodynamic Phase 0 trial of pre-prostatectomy lovastatin to downregulate MYC in localized prostate cancer. | | |
| Role: PI | | |
| 3. PCW Award | Tran (PI) | 4/1/13-3/31/14 |
| Patrick C. Walsh Prostate Cancer Research Fund | | |
| "Phase I Trial of HSP90 Inhibition and Radiation-Androgen Deprivation Therapy for High-Risk Localized and Locally Advanced Prostate Cancer" | | |
| Phase 1 trial of ganetespib and standard of care radiation and long-term androgen deprivation therapy for high-risk and locally advanced prostate cancer. | | |
| Role: PI | | |
| 4. W81XWH-11-1-0336 | Schaeffer (PI) | 11/1/11-9/29/14 |
| Dept of Defense CDMRP Prostate Cancer Research Program | | |
| "RNASEH2A - a Putative "Non-Oncogene Addiction" Gene Target and Marker for Radio-sensitivity in High Risk Prostate Cancer" | | |
| Specific Aim 1: Demonstrate the association of <i>RNASEH2A</i> with lethal prostate cancer. Specific Aim 2: Evaluate the ability of <i>RNASEH2A</i> to modulate radio-sensitivity in prostate cancer cell lines and xenograft models. Specific Aim 3: Investigate <i>RNASEH2A</i> as a tissue and biofluid based marker of radio-sensitivity. | | |
| Role: Co-I | | |

EMPLOYMENT/OPPORTUNITIES:

No employment or research opportunities applied for and/or received based on experience/training specifically from the Aims of this award. However, in the spirit of this award protecting the research time of the PI, this has enabled the PI to be awarded the following opportunities related to prostate cancer research or promote his career:

PROFESSIONAL POSITIONS (Since the beginning of the DoD PRTA):

- | | |
|-----------|--|
| 2010-2013 | Assistant Professor, Oncology (secondary appt), JHU SOM. |
| 2010- | Member, Sidney Kimmel Comprehensive Cancer Center (SKCCC), JHU SOM. |
| 2012- | Member, Graduate Program in Cellular and Molecular Medicine (CMM), JHU SOM. |
| 2013 | Assistant Professor, Urology (secondary appt), JHU SOM. |
| 2013- | Associate Professor, Radiation Oncology, Oncology and Urology, JHU SOM. |
| 2015- | Clinical Director, Radiation Oncology and Molecular Radiation Sciences, JHU SOM. |

AWARDS & HONORS (Since the beginning of the DoD PRTA):

-
- 2012 Association of Residents in Radiation Oncology (ARRO) Educator of the Year.

2012 Top Doctors by Baltimore Magazine.
 2012-2016 American Cancer Society Research Scholar.
 2013 OHSU SOM Alumni Association Early Career Achievement Award (Inaugural Award).
 2013 Alpha Omega Alpha Honor Medical Society Alumni Award, OHSU Chapter.
 2013 The Irene and Bernard L. Schwartz Scholar - Patrick C. Walsh Prostate Cancer Research Fund Award.
 2013-2015 Sidney Kimmel Translational Scholar Award.
 2013-2018 National Cancer Institute (NCI) 1R01CA166348-01A1.
 2015-2016 American Society of Clinical Oncology (ASCO) Leadership Development Program.
 2015-2016 Johns Hopkins Catalyst Award.
 2015-2017 Movember Foundation-Prostate Cancer Foundation (PCF) Challenge Award.

COMMITTEES & PROFESSIONAL ACTIVITIES (Since the beginning of the DoD PRTA):

2011 Co-Chair of JHU SOM Radiation Oncology and Molecular Radiation Sciences *Modulating Radiation Response - Cancer Fundamentals to Therapy* Symposium
 2011-2013 ASTRO Scientific Program Committee - Biology Subcommittee
 2011-2013 NASA Space Radiation Cancer Risks - Ground-Based Studies in Space Radiobiology, Panel Reviewer
 2011-2015 SKCCC Oncology Grand Rounds, Co-Organizer
 2011- SKCCC Service of Remembrance Steering Committee
 2011- SKCCC Educational Committee
 2011- JHU SOM Clinical Practice Association – Compliance Committee
 2011- JHU SOM Clinical Practice Association – Clinical Documentation Excellence Program
 2011- SKCCC Oncology Animal Facility Advisory Committee
 2012 DoD PCRP Clinical and Experimental Therapeutics-2, Ad Hoc Reviewer
 2012 JHU John G. Rangos, Sr., Award for Creativity in Cancer Discovery, Ad Hoc Reviewer
 2012-2013 DoD Prostate Cancer Research Program (PCRP) Pathobiology-1, Scientist Reviewer
 2012-2013 NSCOR - Space Radiation Solid Cancer Risks, Panel Progress Reviewer
 2012-2013 JHU Patrick C. Walsh Prostate Cancer Research Fund, Scientist Reviewer
 2012-2015 ASTRO Radiobiology Practice Exam and Study Guide Committee of the Science Council
 2012-2014 Radiation Oncology Institute National Radiation Oncology Registry (NROR) Pilot Committee
 2012- RSNA Research and Education (R&E) Foundation - Radiation Oncology Research Study Section
 2013 The Halifax Project Task Force: A Broad-Spectrum Integrative Design for Cancer Prevention and Therapy – Genetic Instability Group
 2013 Prostate Cancer UK Pilot Grant, Ad Hoc Reviewer
 2013 DoD PCRP Pathobiology-1, Pre-application Reviewer
 2013-2015 SKCCC Clinical Research Review Committee
 2013-2015 ASTRO Research Grants Evaluation Committee of the Science Council
 2014 NSCOR - Space Radiation Solid Cancer Risks and Biological Countermeasures, Panel Reviewer
 2014-2015 NIH NCI Loan Repayment Program Special Emphasis Panel - ZCA1 PCRB-A (A2) S, Reviewer
 2014-2015 ASTRO NROR Pilot Sites Working Group Committee
 2014-2015 ASTRO Molecular Targeting White Paper Committee
 2014- JHU SOM Instructor/Assistant Professor Reappointment Review Committee
 2014- SKCCC Johns Hopkins-Allegheny Health Network Cancer Research Fund, Co-Director
 2015 RSNA R&E Foundation – Radiation Oncology Research Study Section, Vice Chair
 2015 NIH NCI R03/R21 Program Special Emphasis Panel - ZCA1 SRB-C (M1) S, Reviewer

2015 JHU Patrick C. Walsh Prostate Cancer Research Fund, Scientist Reviewer
 2015 ASTRO Scientific Program Committee - Biology Subcommittee
 2015 Israel Science Foundation, Scientist Reviewer
 2016 Swiss and French Prostate Cancer Foundation-Movember Foundation, Ad Hoc Scientist Reviewer
 2016 Flemish Cancer Society Kom op tegen Kanker (SU2C) , Ad Hoc Scientist Reviewer
 2016-2018 RSNA R&E Foundation – Radiation Oncology Research Study Section, Chair
 2016-2021 NIH NCI RTB R01, Permanent Reviewer

CONFERENCE ORAGANIZER, SESSION CHAIR (Since the beginning of the DoD PRTA):

2011 JHU SOM Radiation Oncology and Molecular Radiation Sciences *Modulating Radiation Response - Cancer Fundamentals to Therapy* Symposium, Co-Chair
 2011 ASTRO National Meeting, Session HH - Nanoparticles and Viruses in Radiotherapy, Co-Chair
 2011 RSNA National Meeting, Radiation Oncology & Radiobiology - Biology, Co-Chair
 2011 RSNA National Meeting, BOOST: Genitourinary – Integrated Science & Practice Session, Co-Chair
 2012 ASTRO National Meeting, Session V - Translational Radiobiology, Co-Chair
 2012-2013 Radiation Research Society (RRS) Annual Meeting Program Committee
 2013 RRS National Meeting, Topical Review – Recent Advancements in Production of Genetically Engineered Mice, Chair
 2013 RRS National Meeting, Symposium – Immune Modulation and Radiation Strategies - Improving Local and Abscopal Responses, Chair
 2013 ASTRO National Meeting, Session F - DNA Damage and Repair: Novel Biological Principles and Targeted Radiosensitization Strategies, Co-Chair
 2013 RSNA National Meeting, Radiation Oncology & Radiobiology - Genitourinary, Co-Chair
 2013 RSNA National Meeting, BOOST: Genitourinary – Integrated Science & Practice Session, Co-Chair
 2014 Prostate Cancer UK 11th Biennial Prostate Cancer Forum – Management: Low-Risk Disease, Co-Chair
 2014 RRS National Meeting, Symposium – Radiation Response of Normal Tissue Stem Cells, Chair
 2014 ASTRO National Meeting, Session SS X - Biology 3 - Biomarkers and Imaging, Co-Chair
 2014 JHU SOM Radiation Oncology and Molecular Radiation Sciences Annual Research Retreat, Co-Organizer
 2015 ASTRO National Meeting, Session EE - Biology V - Imaging and Circulating Biomarkers, Co-Chair
 2016 3rd Amtrak Alliance - Baltimore-Philadelphia Prostate Cancer Summit – Localized Prostate Cancer, Chair
 2016 RRS National Meeting, Symposium – Lung Cancer, Chair
 2017 ASTRO-NCI Immunotherapy and Radiation Oncology Workshop, Co-Chair

EDITORIAL ACTIVITIES (Since the beginning of the DoD PRTA):

2014- Cancer Research, Associate Editor – Breaking Advances

10. REFERENCES:

1. A. Jemal *et al.*, *CA Cancer J Clin* **59**, 225 (Jul-Aug, 2009).
2. C. Kumar-Sinha, S. A. Tomlins, A. M. Chinnaiyan, *Nature reviews* **8**, 497 (Jul, 2008).
3. S. A. Tomlins *et al.*, *Science* **310**, 644 (Oct 28, 2005).

4. V. Rao *et al.*, *Prostate*, (Feb 1, 2012).

11. APPENDIX:

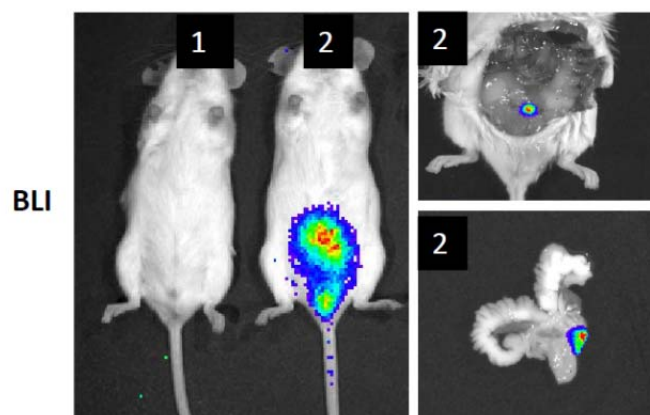


Fig 1. Generation of an inducible luc prostate epithelial specific mouse model. Mice containing a prostate specific TET driver transgene, *Hoxb13-rtTA* was crossed with a reporter mouse *luc-tetO-Twist1* line to produce bi-transgenic animals (HT). The presence of doxycycline allows the rtTA protein to bind and activate the tetO promoter. Removal of doxycycline triggers a conformational change which prevents tetO binding, activation and inhibits Twist and luc transcription. HT animals express luciferase inducibly in the prostate as shown by bioluminescence imaging (BLI) (ip injection with luciferin substrate and imaged 10 minutes later on a Xenogen Spectrum machine shows a colored bright region in the lower abdomen/high pelvis). Dox – doxycycline was given to animals in the drinking water [2 mg/ml]. Animal 1 has a *Hoxb13-rtTA* genotype and animal 2 is an HT mouse. The smaller panels on the right are animal 2 after necropsy and dissection of the prostate and seminal vesicles. In these right panels prostate inducible and specific luc expression can be seen by BLI.

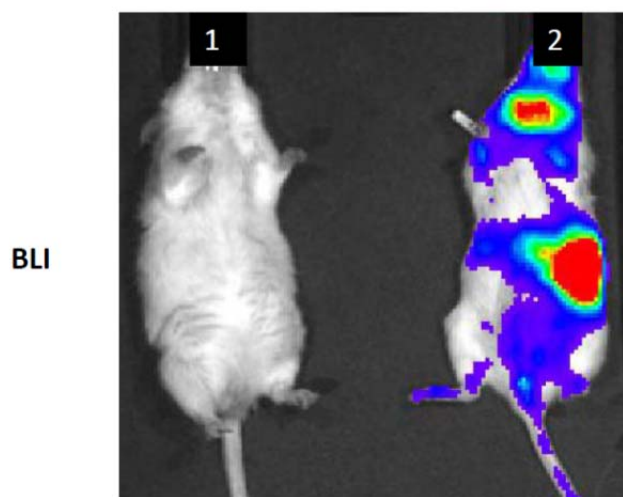


Fig 2. Generation of an inducible luc-tetO-ERG mouse model. Mice containing a ubiquitous TET driver transgene, *CMV-rtTA* were crossed with our ERG line of interest, *luc-tetO-ERG*, that has the luc reporter to generate bi-transgenic animals (CMV-E). The presence of doxycycline allows the rtTA protein to bind and activate the tetO promoter. Removal of doxycycline triggers a conformational change which prevents tetO binding, activation and inhibits ERG and luc transcription. CMV-E animals express luciferase inducibly in the entire mouse as shown by bioluminescence imaging (BLI) (ip injection with luciferin substrate and imaged 10 minutes later on a Xenogen Spectrum machine shows a colored region throughout). Dox – doxycycline was given to animals in the drinking water [2 mg/ml]. Animal 1 has a not been placed on Dox.

Animal Care and Use Committee

1620 McElderry Street
Reed Hall, Room B122
Baltimore, Maryland 21205-1911
(443) 287-3738 / FAX (443) 287-3747
www.jhu.edu/animalcare

Dr. Phuoc Tran
Department of Oncology

Dear Dr. Tran:

On 07/17/2015, the Johns Hopkins University Animal Care and Use Committee (ACUC) approved the following research protocol for which you are the Principal Investigator. A copy of the approved protocol is attached.


Protocol Number: MO15M273

TITLE: Transgenic models of oncogene induced tumorigenesis and organ fibrosis (replaces MO12M261)

The approval period is for three (3) years. The ACUC office will send a notice reminding you to submit the 3rd year replacement protocol. This notice will be sent out 90 days prior to the expiration date. Please use this protocol number when placing an order with Research Animal Resources (RAR) (formerly known as Animal Services). They can be contacted by calling 5-3713. Note: Approval of this protocol does not guarantee University space for housing animals.

You may request modifications to this protocol by submitting the appropriate amendment form (i.e., Change in Animal Number, Change in Personnel, or Change in Procedures) to the ACUC office for review and approval. Copies of all our forms can be found on our website www.jhu.edu/animalcare. For guidance on protocol modifications that require amendments, please refer to the reverse side of this letter. If the locations for outside housing or procedures change, please submit a Change in Location Form, also available on the website.

Sincerely,



Nancy A. Ator, PhD
Chair, Animal Care and Use Committee